

Improving Membrane Filtration for Copper Speciation: Optimal Salt Pretreatments of Polyethersulfone Membranes to Prevent Analyte Retention

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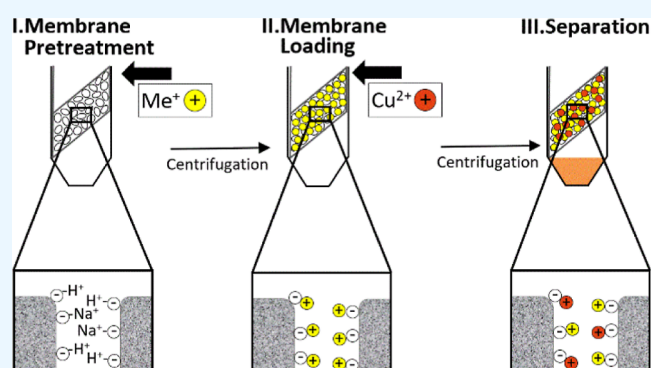
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ABSTRACT: Membrane filtration has been increasingly used to separate dissolved metal ions from dispersed particles, commonly using ultrafiltration membranes, for example, polyethersulfone (PES) membranes with a molecular weight cut-off of 3 kDa. The disadvantage of this technique is an undesired retention of ions, resulting from Coulomb interactions with sulfonic acid groups of the membrane. Therefore, such a membrane acts similar to a cation exchanger column. We solved this drawback by a pretreatment of the PES membrane by other cations. Using CuSO_4 as a model compound, we compared the effectiveness of five cations using their salt solutions (Ca^{2+} , Mg^{2+} , Fe^{2+} , Ag^+ , Ba^{2+}) as pretreatment agents and identified the most effective pretreatment component for a high recovery of copper ions. After membrane filtration without pretreatment, only $52 \pm 10\%$, $64 \pm 5\%$, $75 \pm 8\%$, and $89 \pm 7\%$ of nominal Cu concentrations were obtained using initial concentrations of 0.2, 0.5, 1.0, and 4.0 mg L^{-1} , respectively. The efficiency of the investigated cations increased in the order $\text{Fe} < \text{Ag} < \text{Mg} < \text{Ca} < \text{Ba}$. Furthermore, we analyzed the most efficient concentration of the pretreatment agent. The best performance was achieved using $0.1 \text{ mol L}^{-1} \text{ CaCl}_2$ which increased copper recovery to slightly below 100%, even at the lowest tested Cu concentration (recovery $93 \pm 10\%$ at 0.2 mg L^{-1}). In the environmentally relevant Cu concentration range of 0.2 mg L^{-1} , $0.1 \text{ mol L}^{-1} \text{ BaCl}_2$ was identified as the most efficient pretreatment ($103 \pm 11\%$).



INTRODUCTION

Copper (Cu) is an essential trace element for all living organisms; moreover, in higher concentrations, it has also been an effective agent in biocides and fungicides for decades.^{1,2} Qualitatively meaningful applications reaching the environment are in agriculture (e.g., wine cultivation) as well as in shipping (as an antifouling agent).³ The main targets in these applications are the release of Cu ions and their effectiveness. Consequently, the main exposure pathway of copper ions to the aquatic environment is caused by the release of Cu compounds from antifouling ship paints.

Due to the rapid development of nanotechnology, the use and release of metal-based engineered nanomaterial (ENM) in many fields of application has increased in the past years. A drawback of the innovative and dynamic technology of ENM is the disposal of the newly designed products and particles without adequate knowledge of their properties and behavior into the environment. In recent years, a large number of publications has pointed out a risk of ENM to aquatic and terrestrial organisms.^{2,4,5} For further application of these innovative materials, it is important to know the cause of the main effects. There are different hypotheses based on the enhanced ion release or effects of the ENM itself.^{6,7} This is

especially the case for copper oxide nanoparticles (CuONPs) with solubility and electroactivity that highly depend on solution composition.⁸ Therefore, it is of major importance to use reliable methods in the analysis of environmental samples to distinguish nanoparticulate copper from dissolved copper, that is, to assess the solubility, the ion release, and the total content of the ENM.

Well-established methods for the determination of copper concentrations in aqueous samples are titration methods like iodometry,⁹ colorimetry,¹⁰ ion chromatography,¹¹ or flame atomic absorption spectrometry.¹² These methods work well down to the low mg L^{-1} concentration range.

There are also electrochemical methods like electrogravimetry or the use of ion-selective electrodes. The latter can be used for the determination of dissolved copper, but the applicability for environmental samples is low because it works

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reliably only in a low pH range ($\text{pH} < 6$) and at low concentrations of halides (Cl^- , Br^- and I^-) or other Cu^{2+} complexing substances that interfere.¹³

For lower copper concentrations down to the low $\mu\text{g L}^{-1}$ range, inductively coupled plasma optical emission spectrometry and mass spectrometry (ICP–OES/MS) or graphite furnace atomic absorption spectrometry (GF–AAS) are common methods for the determination.^{14–17}

Alternatively, the dissolved fraction of dispersed nanoparticles can be determined with separation methods like the well-established membrane filtration using ultrafiltration (UF) membranes or ultracentrifugation (UC).¹⁸ Newer approaches for this are single-particle (sp) ICP–MS or the use of field-flow fractionation methods ideally coupled with an ICP–MS detector.^{14,19} Membrane filtration and UC are easily available methods, considering the facile use of filtration devices (namely ready-to-use centrifugal UF devices, for example, Sartorius Vivaspin, Merck Amicon, or Pall–Gelman Microsep) and the good availability of efficient centrifuges. We focus in the following on these two easily available separation methods.

A reliable separation method for the determination of the dissolved fraction of ENM should have a low size cut-off in the very low nanometer range, preferably at ≈ 1 nm. Additionally, it should be fast enough to correctly assess the even faster dissolution kinetics of ENM in preferably hourly steps.^{20–22}

In this context, an important advantage of the membrane filtration is that a relatively short time is needed for the separation step. Considering the nanomaterial CuO we have focused on, the membrane UF separation protocol used here requires 30 min to achieve a diameter cut-off of < 1.2 nm [with PES, molecular weight cut-off (MWCO) 3 kDa, see the [Experimental Section](#)]. Conventional separation by centrifugation with an Eppendorf 5430 R centrifuge with maximal settings (30,130g) resulted in a much larger cut-off diameter of ≈ 8 nm for CuONPs in water after 30 min of centrifugation.²³ In order to achieve a UF comparable cut-off diameter of < 1.2 nm with this centrifuge (Eppendorf 5430 R), a centrifugation duration of far more than 8 h would be required; even with an ultracentrifuge with 390,000g, the duration would be approx. 6 h. The correct assessment of faster dissolution kinetics of small nanoparticles would be very limited with such long separation times. Therefore, we further limited our study to UF as the separation method.

Typical membrane materials of centrifugal UF devices are regenerated cellulose (RC) (Amicon Ultracel, Vivaspin Hydrosart), cellulose triacetate (Vivaspin CTA), or polyethersulfone (Vivaspin PES or Pall Omega). These devices are commercially available with membranes down to MWCO values of 1 kDa (PES, Pall Omega), 2 kDa (RC, Sartorius Hydrosart), and 10 kDa (CTA, Sartorius), respectively.

Other typical membrane materials used for UF are, for example, polyvinylidene fluoride, polytetrafluoroethylene, polysulfone, polyamide, and polyacrylonitrile.²⁴ However, ready-to-use commercially available centrifugal filtration devices using these membrane materials provide only size cut-offs down to $0.1 \mu\text{m}$.

In the past decades, membrane filtration has been increasingly used to separate dissolved metal ions from dispersed particles, in fact often using PES or RC membranes.^{18,21,25–28} A direct comparison of the three types of membrane materials, PES, cellulose triacetate (CTA), and Hydrosart (RC) at MWCO 10 kDa, shows the highest water flux, the widest pH compatibility, and the shortest separation

duration (e.g., bovine serum albumin) for the PES-based centrifugal UF devices.²⁹ Hence, we focused on PES-based UF devices.

Microporous PES membranes are commonly produced from PES polymers using the Loeb–Sourirajan phase inversion method.^{24,30,31} The PES polymers ([Figure 1](#)) typically contain

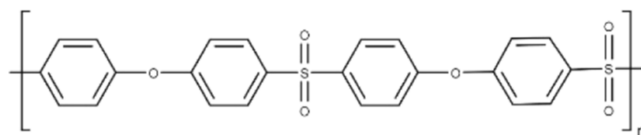


Figure 1. Chemical structure of a polyethersulfone polymer.

a small ratio of free sulfonic acid groups directly from the synthesis using compounds containing sulfonic acid groups³² or from subsequent modification of the membrane to achieve increased hydrophilicity by introducing additional sulfonic acid groups.²⁴

Consequently, these strongly acidic sulfonic acid groups are deprotonated and negatively charged at neutral pH. Hence, the zeta potential of the PES membrane is typically strongly negative as reported by Pöhler et al. with approx. -40 mV in the pH range 8–10 and increases toward isoelectricity for a decreasing pH, almost reaching the isoelectric point at pH 2.³³ The negatively charged groups of the membrane interact with the positively charged cations (e.g., the Cu^{2+} cations) and therefore may cause an undesired retention of cations by Coulomb interaction, which was also reported for several other common filtration membrane materials.³⁴ Apparently, divalent or trivalent cations exhibit stronger electrostatic interactions with the negative surface charges.

The presence of complexing substances in the feed, like ethylenediaminetetraacetic acid (EDTA), has a strong influence on the speciation of the cations, for example, the formation of the negatively charged complex $[\text{CuEDTA}]^{2-}$ with Cu^{2+} , thus supposedly impeding the attractive Coulomb forces. Otherwise, Weltje et al. and Salbu et al. hypothesized that negatively charged EDTA complexes may have been the cause for the affinity of di- or trivalent metals to polyester filters.^{34,35} The transport through UF membranes with a size cut-off ≥ 1 nm operates exclusively with the pore-flow mechanism and is accomplished via a size-sieving mechanism.²⁴ Nanofiltration membranes with a size cut-off ≤ 1 nm (and MWCO ≤ 1 kDa) show a combination of solution-diffusion and pore-flow character.²⁴ In this size cut-off range, the polymer surface charge may exhibit a low to moderate rejection of higher valent ions and low rejection of monovalent ions.²⁴ Therefore, we chose a centrifugal UF device using a PES membrane with a slightly larger MWCO of 3 kDa and expected predominantly pore-flow mechanism and only low rejection of ions due to membrane surface charge effects.

Considering the typical complex porous structure of PES membranes and its sulfonic acid groups on the surface (see [Figure 2](#)), which are common also for strongly acidic cation exchangers,³⁶ the filtration membrane could therefore act similar to a small cation exchange column only with a low ion exchange capacity (IEC).^{31,34,37}

The key challenge of undesired retention during membrane filtration was addressed by Cornelis et al. in the context of the investigation of the retention of silver and cerium oxide nanoparticles in soils and the determination of the dissolved fraction after micro- and UF.²⁵ They found that the

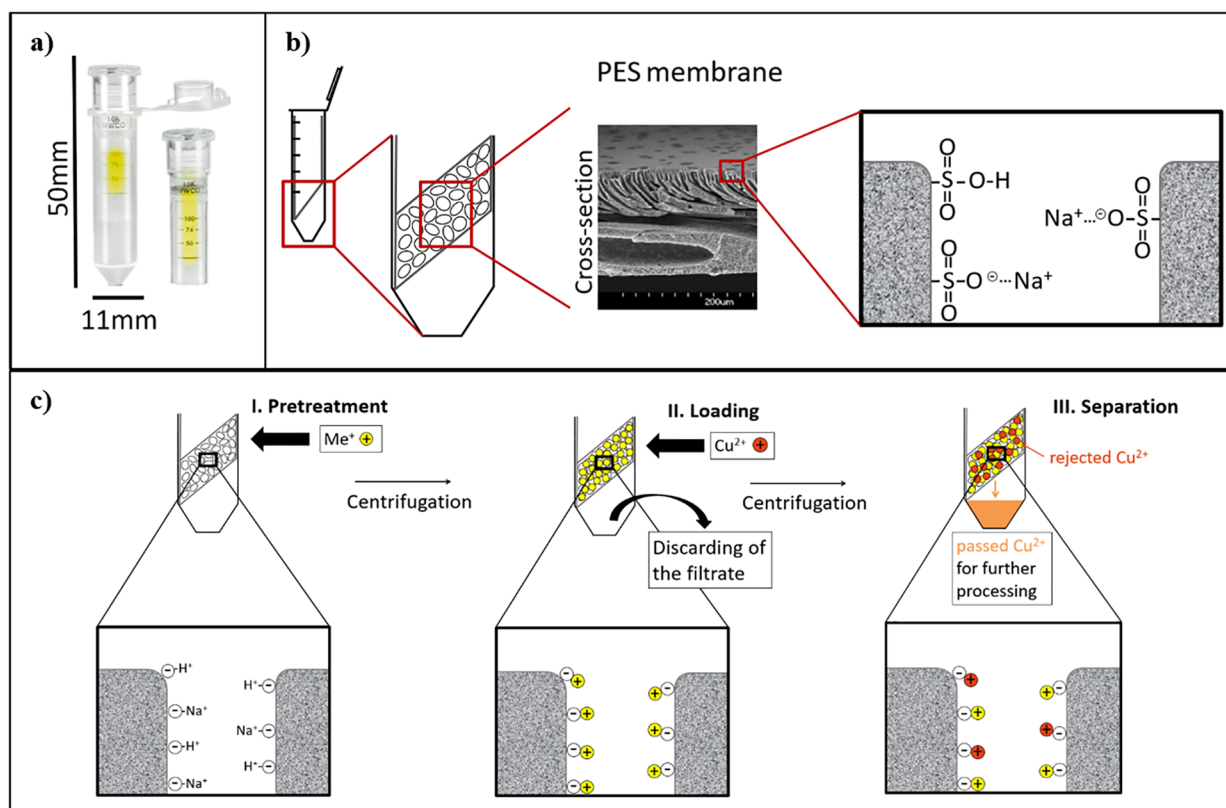


Figure 2. (a) Filtrate vessel and centrifugal filter unit with PES membrane filter, (b) morphology of the PES membrane, scanning electron microscopy modified according to Son et al.³⁰ (with courtesy of Son et al. Copyright 2022), and (c) schematic of the procedure for improving membrane UF: (I) pretreatment of the PES membrane with metal cations (e.g. Ca^{2+} , Mg^{2+} , Ag^+); (II) loading of the centrifugal filter unit (PES membrane) with Cu^{2+} ; and (III) separation of the dissolved Cu fraction by centrifugation of the centrifugal UF device.

pretreatment of the used filters with $\text{Cu}(\text{NO}_3)_2$ solution (0.1 mol L^{-1}) increased the recoveries of dissolved Ag and Ce^{III} and attributed this to Cu^{2+} occupying specific binding sites on the membranes by reversible and hence transient interactions, thus preventing subsequent Ag^+ and Ce^{3+} adsorption. Therefore, they chose the use of filters with this Cu pretreatment as the preferred separation method for Ag- and Ce-containing solutions. This approach has been widely used and is accepted for UF in studies on silver and silver sulfide nanomaterials.^{38–41} However, a similar approach to prevent the retention of Cu^{2+} when using UF has not been published so far.

Preliminary experiments in our lab using UF for the determination of the dissolved fraction of Cu were conducted with CuCl_2 solutions in a copper concentration range from 0.2 up to 4.0 mg L^{-1} ($3.15\text{--}62.95 \text{ } \mu\text{mol L}^{-1}$) using centrifugal UF devices with PES membranes (for details, see [Supporting Information](#)). This concentration range was chosen to cover the reported effective concentration values from ecotoxicity studies using crustaceans, fish, algae, nematodes, protozoa, bacteria, and yeast.^{42–44} The initial results clearly indicated retention of copper ions by the PES membrane (see [Figure S1](#)). At 4.0 mg L^{-1} , only a negligible fraction of copper ions was retained. However, with decreasing Cu concentration, the fraction of retained Cu increased strongly.

A previous study focusing on silver nanoparticle speciation in different artificial aqueous media compared results from UC and UF (also with PES membranes).¹⁸ All studied media showed higher dissolved silver content when using UC, with the exception of the standard freshwater medium Elendt M7.⁴⁵

Therefore, we considered the possibility that the major divalent cations of the test medium Ca^{2+} and Mg^{2+} could have prevented the retention of Ag^+ in these UF experiments.

The aim of this study was therefore to prevent the retention of Cu ions when using UF. By adapting the approach of Cornelis et al.²⁵ and considering the peculiarities of Cu ions, we used cations that have even stronger interactions with the membrane. The cation affinities on sulfonated polystyrene ion exchange resins suggest, for example, Ca^{2+} , Ag^+ , or Ba^{2+} with stronger affinities compared to Cu^{2+} as potential pretreatments.⁴⁶

We evaluated Cu solutions in the concentration range up to 4.0 mg L^{-1} and used UF with a PES membrane. According to the assumptions above, we tested solutions of Ca^{2+} and Mg^{2+} as well as Ag^+ and Ba^{2+} solutions for the membrane pretreatment and finally an Fe^{2+} solution at the concentration of the standard freshwater medium Elendt M7.

The retention of the membrane was determined by comparing the Cu concentration of the filtrates with the total Cu concentration for the different pretreatments. The measurement of the Cu concentration was done by GF-AAS after digestion.

The objectives of this study were (i) identification of the most effective pretreatment component for a high recovery (and low retention) of Cu ions and (ii) identification of the most effective concentration of this component for the pretreatment.

EXPERIMENTAL SECTION

Materials. The pretreatment solutions were prepared with double-distilled water (ddH₂O) using the salts magnesium sulfate (MgSO₄·7H₂O), calcium chloride (CaCl₂·2H₂O), ferrous sulfate (FeSO₄·7H₂O), and EDTA disodium salt dihydrate (EDTA-Na₂) that were purchased from Merck KGaA (Darmstadt, Germany). Additionally, barium chloride (BaCl₂·2H₂O) was purchased from Alfa Aesar GmbH & Co. KG (Karlsruhe, Germany), and silver nitrate (AgNO₃) was purchased from Carl Roth GmbH & Co. KG (Arlenheim, Germany).

The copper salt solutions for the determination of membrane retention were prepared with copper sulfate (CuSO₄·5H₂O) obtained from Merck KGaA (Darmstadt, Germany) and diluted with ddH₂O to achieve the tested concentrations. The pH of the solutions was measured using a pH meter (ProLab 2500, SI Analytics, Weilheim, Germany, equipped with the electrode SenTix 41, WTW, Weilheim, Germany). For Cu concentrations 0.2 and 1.0 mg L⁻¹, the pH was controlled to be 7.1 and 7.0, respectively; therefore, we straightforwardly assumed neutral pH 7 for the prepared Cu solutions.

For sample digestion, nitric acid (HNO₃, puriss p.a., ≥99%, 65%) and hydrogen peroxide (H₂O₂, p.a., 35%) from Sigma-Aldrich GmbH (Steinheim, Germany) were purchased. For the GF-AAS measurements, copper standard solution (1000 mg L⁻¹, traceable to SRM from NIST Cu(NO₃)₂ in HNO₃ 0.5 mol L⁻¹, CertiPUR) and palladium nitrate matrix modifier solution (Pd(NO₃)₂, 10.0 ± 0.2 g L⁻¹, in 15% HNO₃) from Merck KGaA (Darmstadt, Germany) were used.

Sample preparation including storage, digestion, and dilution was done using disposable tubes obtained from Sarstedt [50 mL, polypropylene (PP), Sarstedt, Nümbrecht, Germany] and Eppendorf (1.5 and 2.0 mL, PP, Eppendorf, Hamburg, Germany). Pipette tips (2–200 μL, 50–1000 μL, and 0.1–5.0 mL, PP) and adjustable-volume pipettes (Eppendorf Research Plus, 20–200 μL, 100–1000 μL, and 0.5–5.0 mL) were also purchased from Eppendorf. Sample cups (PP) for the autosampler were obtained from Thermo Fisher Scientific (Dreieich, Germany).

Determination of Cu Content by GF-AAS. Cu concentrations were quantified by GF-AAS following Scheiber et al.¹⁶ using a Unicam 989 QZ AA spectrometer (Unicam, Cambridge, UK) with a GF-90 plus furnace and a FS-90 plus autosampler. The device was controlled using the software Solaar32 Data Station. The calibration of the GF-AAS was performed in the concentration range of 0–200 μg L⁻¹ using the appropriate dilution of the above-mentioned CertiPUR Cu standard solution (1000 mg L⁻¹).

All samples were digested before GF-AAS measurement by adding 50 μL of H₂O₂ (35%) and 50 μL of HNO₃ (65%) to a sample volume of 100 μL. After heating (85 °C) overnight to dryness, the sample was dissolved in 1000 μL of diluted HNO₃ (1%) and then measured by GF-AAS. In case the estimated Cu concentration was higher than the upper limit of the working range (200 μg L⁻¹), subsequent dilution steps using diluted HNO₃ (1%) were performed until the working range was reached (for details, see Supporting Information).

UF of Cu Solutions. UF was done using centrifugal UF devices Vivaspin 500 containing PES membranes with a MWCO of 3 kDa (MWCO 3000, Sartorius, Göttingen, Germany). These devices consist of a filtrate vessel (2.0 mL,

PP) holding a removable centrifugal filter unit (PC) with the membrane (PES) fixed inside it (see Figure 2a). The active membrane area is 0.5 cm².²⁹ The pore size of the membrane was estimated to be 1.2 nm.⁴⁷ The filtration of Cu solutions was done by placing 500 μL of the sample in the centrifugal filter unit before centrifuging for 30 min at ≈14,000g (MiniSpin plus, Eppendorf, Hamburg, Germany). Then, a volume of 100 μL of the filtrate was transferred to a 1.5 mL tube and digested.

The Cu content was subsequently measured by GF-AAS. For the direct comparison in each set of measurements, a volume of 100 μL of the used Cu solution was also digested and subsequently measured by GF-AAS.

Membrane Pretreatment. For the membrane pretreatment, 500 μL of the respective pretreatment solution (Table 1) was placed in the centrifugal filter unit with the membrane (inside the 2.0 mL filtrate vessel holding the unit). The filtration devices were centrifuged for 30 min at ≈14,000g (using the Mini-Spin plus centrifuge). Afterward, the filtrate vessel was disposed, as well as the filtrate. Potential residual feed on the membrane inside the centrifugal filter unit was removed by a pipette with a small tip (2–200 μL, PP) and also discharged. The centrifugal filter unit was put into a new tube (PP, 2.0 mL, with the lid removed). Then, the centrifugal filter unit was immediately used for the next step to avoid dehydration of the membrane as recommended by the supplier.⁴⁸

Table 1. Solutions Used for Membrane Pretreatments

pretreatment label	membrane pretreatment solution
UF (Mg ²⁺)	MgSO ₄ 0.5 mmol L ⁻¹
UF (Ca ²⁺)	CaCl ₂ 2.0 mmol L ⁻¹
UF (Ca ²⁺ & Mg ²⁺)	CaCl ₂ 2.0 mmol L ⁻¹ MgSO ₄ 0.5 mmol L ⁻¹
UF (Fe ²⁺ & EDTA)	FeSO ₄ 3.6 μmol L ⁻¹ EDTA-Na ₂ 6.7 μmol L ⁻¹
UF (Ag ⁺)	AgNO ₃ 0.59 mmol L ⁻¹
UF (Ca ²⁺ & Mg ²⁺ 2.4 mM)	CaCl ₂ 2.0 mmol L ⁻¹ MgSO ₄ 2.4 mmol L ⁻¹
UF (Ca ²⁺ 0.1 M)	CaCl ₂ 0.1 mol L ⁻¹
UF (Ba ²⁺ 0.1 M)	BaCl ₂ 0.1 mol L ⁻¹

Subsequently, 500 μL of the tested Cu solution was placed in the centrifugal filter unit (inside the new 2.0 mL tube holding the unit) and was once again centrifuged for 30 min at ≈14,000g (see Figure 2c). Thereafter, the centrifugal filter unit was discarded, and the Cu concentration of the filtrate was measured by GF-AAS after digestion.

The membranes themselves were not analyzed because the removal of the membranes from the very small centrifugal filter units was not feasible without severe damage and contamination of the membranes.

Data Evaluation and Statistical Analysis. Statistical analysis was performed using R.⁴⁹ Mean and median values, standard deviation, interquartile range, and confidence interval (ci) were calculated per treatment. All data were analyzed for significant differences between pretreatments at the different concentrations. Comparison was performed by using a nonparametric Wilcoxon test or Kruskal–Wallis test and a Benjamini–Hochberg⁵⁰ post-hoc test.

The error propagation was calculated using Mathematica and Matlab (for details, see Supporting Information).

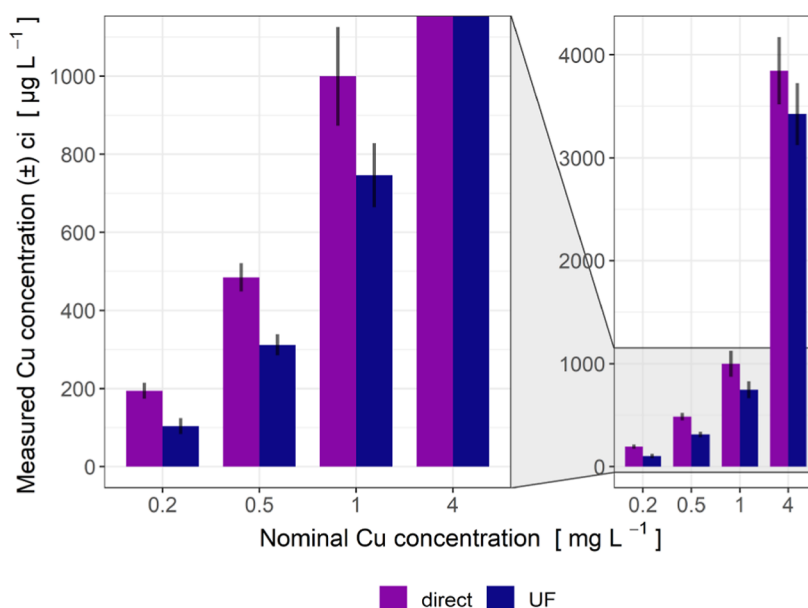


Figure 3. Measured Cu concentrations (mean \pm ci) of solutions with different nominal Cu concentrations of 0.2, 0.5, 1.0, and 4.0 mg L⁻¹ without (light violet bars, $n \geq 15$) and with UF (dark violet bars, $n \geq 10$).

RESULTS AND DISCUSSION

UF without Pretreatment. The measured Cu concentrations of the CuSO₄ solutions (0.2–4.0 mg L⁻¹) without filtration are very well in range of the nominal Cu concentrations (light violet bars, see Figure 3). The results for the filtrated samples using UF showed a significantly reduced recovery ($p < 0.01$) of Cu for the solutions with the three low nominal Cu concentrations 0.2, 0.5, and 1.0 mg L⁻¹ compared to measurement without UF (dark violet bars, Figure 3). With the increasing nominal concentration, the determined Cu recovery after UF also increased. For the lowest nominal Cu concentration of 0.2 mg L⁻¹, the recovery was only 0.104 ± 0.018 mg L⁻¹ ($53 \pm 10\%$). For the higher Cu concentration of 0.5 and 1 mg L⁻¹, the Cu recovery was 0.312 ± 0.017 mg L⁻¹ ($64 \pm 6\%$) and 0.747 ± 0.082 mg L⁻¹ ($75 \pm 10\%$), respectively, after UF. For the highest nominal Cu concentration, the recovery was 3.424 ± 0.247 mg L⁻¹ ($89 \pm 8\%$) after UF.

These values confirm our results from preliminary UF experiments, and similar reduced recoveries were reported for other membranes and cations, for example, Ag⁺, Ce³⁺, Zn²⁺, and Mn²⁺ in the concentration range below 1 mg L⁻¹.^{25,26,34} As proposed above, we assumed that this reduction was caused by Coulomb interaction of the positively charged Cu²⁺ cations with the negatively charged sulfonic acid groups of the PES membrane.

Based on the retained Cu amounts in the range of 45–210 ng per membrane (0.71–3.30 nmol) (shown in the Supporting Information, Figure S2), we were able to estimate the IEC of the centrifugal filter units of $0.24\text{--}0.66$ $\mu\text{equiv g}^{-1}$ (omitting the highest retention value due to its large ci). The ci values of the UF for Cu concentration of 1.0 mg L⁻¹ were still in the range of the data itself, so we did not discuss a possible trend of the data. The estimated IEC range was in comparison lower by a factor of approximately 1000 than typical values for sulfonated PES ion exchange membranes (0.4–1.6 mequiv g⁻¹) and also lower than that for unmodified PES membranes (0.03–0.08 mequiv g⁻¹).^{31,51} The retention of the PES

membrane of centrifugal filter units is generally very low (e.g., for proteins 2–10 $\mu\text{g cm}^{-2}$) and may increase in case the filtrate is of interest (for proteins up to 20–100 $\mu\text{g cm}^{-2}$).⁵² For the small protein aprotinin (6.5 kDa), this would be 1.5–7.7 nmol for a membrane area of 0.5 cm². This amount would be in the same molar range we observed for the much smaller cation Cu²⁺, even if the adsorption of proteins to PES membranes is far more complex.⁵³ However, if very low concentrations of toxic metal ions like Cu²⁺ in $\mu\text{g L}^{-1}$ range (e.g. up to 500 $\mu\text{g L}^{-1}$) are of interest, the observed membrane retention may strongly bias the assessment of the environmental risk.

To reduce the interaction between the membrane and the Cu ions affecting the quantification, the filter membrane was pretreated following Cornelis et al.²⁵ The cations in the proposed pretreatment solutions should interact with the sulfonic acid groups of the PES membrane more strongly compared to Cu ions. Consequently, the Cu ions are displaced and pass the membrane during UF.

Identification of the Most Effective Pretreatment.

Assuming that the interaction of the PES membrane especially with bivalent cations increases the recovery of Cu from the used CuSO₄ test solutions, the most promising cations of the Elendt M7 medium (Table 1) were tested as pretreatment (see Figure 4). Following the considerations of the Introduction Section, the concentrations of the first set of pretreatment solutions corresponded to those in the technical guideline for the Elendt M7 medium and were in the low mmol L⁻¹ (Ca²⁺, Mg²⁺, Ag⁺) and low $\mu\text{mol L}^{-1}$ range (Fe²⁺ and EDTA).⁴⁵ Considering the tested Cu concentration range of 3.15–62.95 $\mu\text{mol L}^{-1}$, the pretreatment concentrations were in large excess for the cations Ca²⁺, Mg²⁺, and Ag⁺ and for the cation Fe²⁺ with EDTA in the range of the lower Cu concentration (see Table 1).

The first series of experiments were conducted using CuSO₄ solutions with the nominal Cu concentration of 1.0 mg L⁻¹ (15.74 $\mu\text{mol L}^{-1}$). For the pretreatment with MgSO₄ (0.5 mmol L⁻¹) and CaCl₂ (2.0 mmol L⁻¹), copper recovery of only 0.538 ± 0.107 mg L⁻¹ ($54 \pm 11\%$) and 0.596 ± 0.128 mg L⁻¹

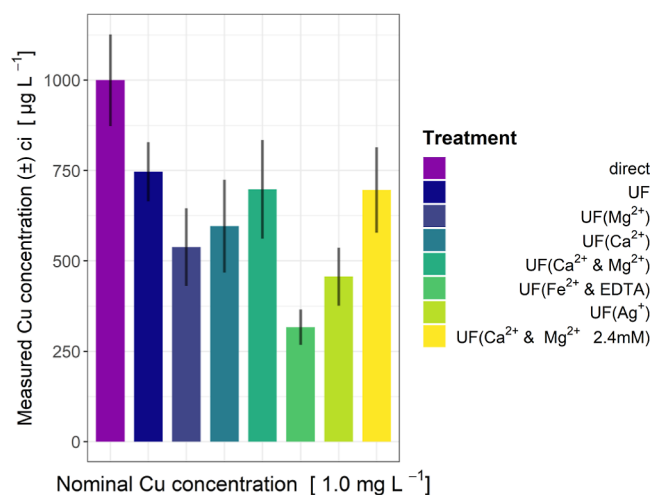


Figure 4. Measured Cu concentrations of solutions with a nominal Cu concentration of 1.0 mg L^{-1} after membrane filtration with different pretreatments ($n \geq 3$; mean \pm ci) in comparison to the direct measurement without UF (light violet bar). Concentrations of the pretreatments are listed in Table 1.

($60 \pm 13\%$), respectively, could be achieved. Both these results were slightly lower than for UF without pretreatment ($0.747 \pm 0.082 \text{ mg L}^{-1}$; $75 \pm 8\%$). Based on the typical series of relative affinity of cations on sulfonated polystyrene cation exchange resins of $\text{Na}^+ < \text{Mg}^{2+} < \text{Cu}^{2+} < \text{Ca}^{2+} < \text{Ag}^+ < \text{Ba}^{2+}$, we expected a slightly lower recovery after the Mg^{2+} pretreatment and a higher recovery after Ca^{2+} pretreatment in comparison to the UF without pretreatment.⁴⁶ Contrary to this generally accepted series, our measurements would indicate a stronger affinity of Cu^{2+} to the PES membrane compared to Ca^{2+} and Mg^{2+} .

However, we suppose an effect of the trace amount of sodium azide (NaN_3) that is used as a preservative in the membrane.⁵⁴ The azide anion N_3^- forms complexes $[\text{Cu}$

$(\text{N}_3)_n]^{2-n}$ with Cu^{2+} and would therefore reduce the affinity of Cu^{2+} to the negatively charged sulfonic acid groups of the PES membrane in the beginning of the filtration process until the preservative NaN_3 is flushed out of the PES membrane.⁵⁵ This would slightly increase the recovery of Cu^{2+} . In fact, the comparison of our results for the UF and the UF with a rinsing step with ddH₂O supports this assumption (see Figure S4). In case of the pretreated membranes (with Mg^{2+} or Ca^{2+}), the preservative NaN_3 would have been flushed out and would not increase the recovery; only the cations Mg^{2+} and Ca^{2+} would influence the recovery of Cu^{2+} during UF.

The combination of the most promising cations (Ca^{2+} and Mg^{2+}) at the above used concentrations slightly increased the recovery to $0.698 \pm 0.137 \text{ mg L}^{-1}$ ($70 \pm 14\%$) (see Figure 4). Increasing the Mg^{2+} concentration to 2.4 mmol L^{-1} in the combination with Ca^{2+} (2.0 mmol L^{-1}) in the pretreatment resulted in an unchanged Cu recovery of $0.696 \pm 0.118 \text{ mg L}^{-1}$ ($70 \pm 12\%$). This small increase was expected but was not significantly different to the UF without pretreatment.

The pretreatment with Ag^+ (0.59 mmol L^{-1}) resulted in a significant reduced recovery ($p < 0.001$) of $0.457 \pm 0.080 \text{ mg L}^{-1}$ ($46 \pm 8\%$) compared to UF without pretreatment. This suggested a weaker affinity of Ag^+ to the membrane compared to Cu^{2+} contrary to the expected affinity series shown above but would be in line with the trend of Coulomb interactions.

The pretreatment with Fe^{2+} ($3.6 \mu\text{mol L}^{-1}$) and EDTA- Na_2 ($6.7 \mu\text{mol L}^{-1}$, used for stabilization of Fe^{2+} in the Elenit M7 medium) did not produce promising results. The determined recovery was even lower with only $0.317 \pm 0.049 \text{ mg L}^{-1}$ ($32 \pm 5\%$) (Figure 4). It was likely that the formation of very stable complexes, like $[\text{Fe}(\text{EDTA})]^{2-}$, led to a strongly reduced affinity to the PES membrane and therefore Fe^{2+} was not available for the blocking of possible binding sites of the PES membrane.

Additionally, a rinsing step after the Fe^{2+} and EDTA pretreatment was tested for a subset of samples for removing the remaining ions which were not adsorbed. The rinsing step

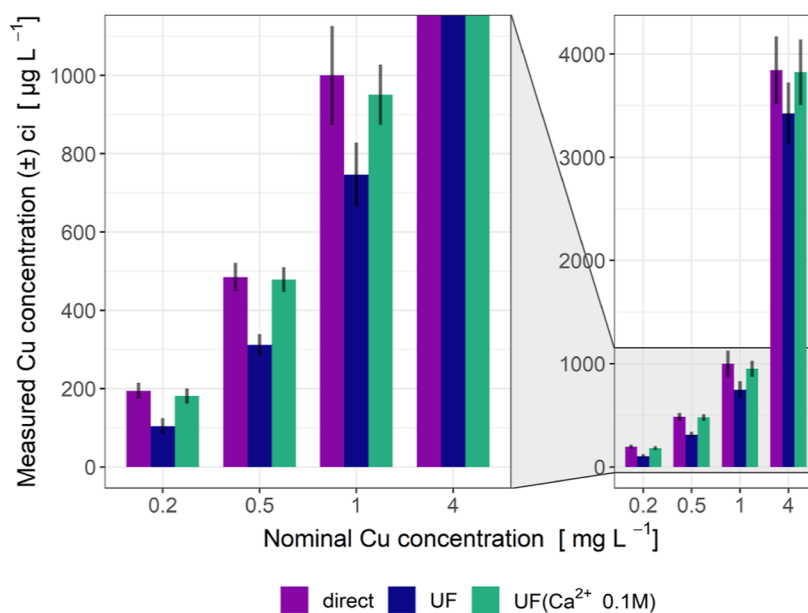


Figure 5. Measured Cu concentrations of solutions with different nominal Cu concentrations of 0.2 , 0.5 , 1.0 , and 4.0 mg L^{-1} after UF without pretreatment (dark violet bars, $n \geq 10$) and with 0.1 M CaCl_2 pretreatment of the filter membrane (green bars, $n \geq 6$, mean \pm ci) in comparison to the direct measurement without UF (light violet bars).

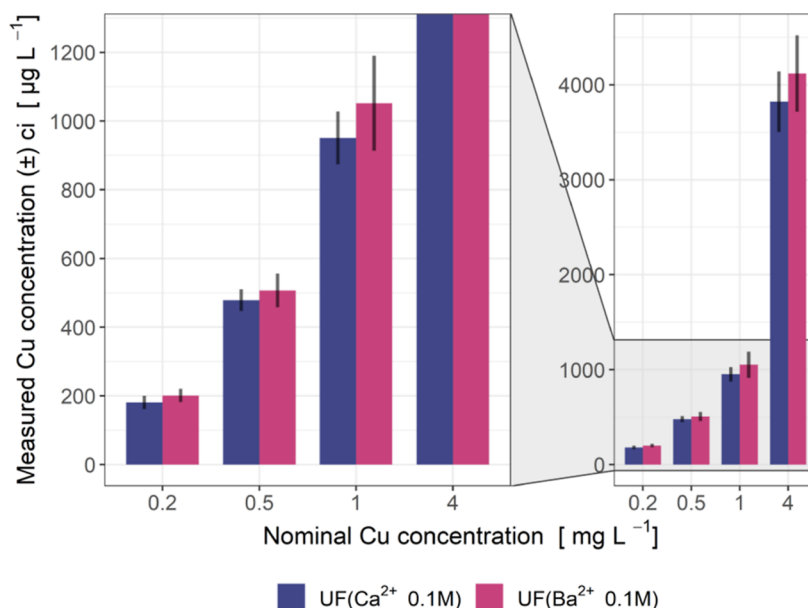


Figure 6. Measured Cu concentrations of solutions with the nominal Cu concentrations 0.2, 0.5, 1.0, and 4.0 mg L⁻¹ after UF with pretreatment 0.1 mol L⁻¹ CaCl₂ (dark violet bars, $n \geq 6$; mean \pm ci) in comparison to the pretreatment with 0.1 mol L⁻¹ BaCl₂ (pink bars, $n \geq 5$, mean \pm ci).

further reduced the recovery to 0.184 ± 0.036 mg L⁻¹ (18 \pm 4%) (see Figure S5a). This may have been caused by a further washing out of the remaining ions off the membrane. Even less recovery of 0.131 ± 0.015 mg L⁻¹ (13 \pm 2%) was determined when only Fe²⁺ and no EDTA was used (see Supporting Information, Figure S5a). This may have been caused by the likely oxidation of Fe²⁺ to Fe³⁺ by atmospheric O₂ and subsequent precipitation of hydrous ferric oxides (e.g., Fe(OH)₃) on the membrane, that would provide additional binding sites for the sorption of Cu²⁺ on the membrane.^{56,57} The oxidation may have also happened in the case of Fe²⁺ and EDTA. However, EDTA also stabilizes the Fe³⁺ cation in solution by the formation of the complexes [Fe(EDTA)]⁻ and [FeOH(EDTA)]²⁻.

Based on the trend of the recovered Cu concentrations so far and ignoring the possible influence of NaN₃, we suppose an affinity series of Fe²⁺ and EDTA < Ag⁺ < Mg²⁺ \approx Ca²⁺ \approx Cu²⁺. This would strictly follow the trend of Coulomb interaction with the sulfonic acid groups of the membrane, considering also the proposed formation of Fe–EDTA complexes.

This sequence would be in accordance with the reported retention times of Cu²⁺ by Nordmeyer et al.⁵⁸ They obtained retention times close to Ca²⁺ for transition-metal cations except for Cu²⁺, which showed broad peaks and variably longer retention times than Ca²⁺ depending on the eluent composition. Therefore, the use of the pretreatments in the concentration of Elendt M7 was not successful in achieving higher Cu recoveries for UF. At these concentrations, the pretreatment before UF of Ag⁺ solutions might still work because Ag⁺ has a weaker interaction with the sulfonic acid groups. Additionally, we suppose equilibrium conditions for the cation interactions with the UF membrane according to the literature on ion exchange processes.^{31,34,37}

Concluding from these results and the data from Nordmeyer et al.,⁵⁸ the use of CaCl₂ was identified as the most promising pretreatment of the membranes so far.

Pretreatment with Divalent Cations Ca²⁺ and Ba²⁺ in Large Excess at 0.1 mol L⁻¹. For the next measurement series, the concentration of Ca²⁺ was increased to an even

larger excess of 0.1 mol L⁻¹ like the proposed concentration for the pretreatment in the work of Cornelis et al.²⁵

Overall, Cu recovery was improved in all tested concentrations by pretreating the UF membrane with 0.1 mol L⁻¹ CaCl₂ (Figure 5). There was some variation in recovery, depending on the concentration of the Cu solution. For the higher Cu concentrations of 0.5 to 4 mg L⁻¹, the recovery was acceptable with values in the range of 95–99%. For the lowest concentration of 0.2 mg Cu L⁻¹, a slightly lower recovery of 0.181 ± 0.019 mg L⁻¹ (93 \pm 10%) was achieved. This steep increase in the recovery of Cu compared to the pretreatments in the mmol L⁻¹ range in the previous section was attributed to the now applied large excess of Ca²⁺ in the pretreatment, which effectively prevented the retention of Cu²⁺ interacting with the PES membrane. An additional effect could arise from the elevated ionic strength, which, according to the Debye–Hückel theory, shields the Coulomb interaction of the Cu ions with the membrane. This could have reduced the retention by the membrane. Due to the fact that the sample itself washes some of the CaCl₂ pretreatment out of the membrane, the effective concentration in the membrane could have been lower than 0.1 mol L⁻¹ but high enough to displace the Cu ions during UF.

Based on the fact that environmentally relevant concentrations are mainly found in lower concentration ranges, another cation (Ba²⁺ as BaCl₂) was tested as pretreatment to further improve the recovery at low Cu concentrations. Considering the above-mentioned affinity series of cations on sulfonated polystyrene cation exchange resins⁴⁶ and the classical data of Zenki,³⁷ it was obvious to assume that the binding strength of Ba²⁺ to the sulfonic acid groups of the membrane is stronger than that of Ca²⁺ and therefore Ba²⁺ should well maintain the binding to the membrane in competition with Cu ions.

On pretreating the UF membrane with 0.1 mol L⁻¹ BaCl₂, the Cu recovery was even higher than with the pretreatment using 0.1 mol L⁻¹ CaCl₂. Figure 6 compares the Cu recovery for the Cu concentration range of 0.2–4.0 mg L⁻¹ with these pretreatments. The Cu recovery was slightly above 100% with

$0.201 \pm 0.019 \text{ mg L}^{-1}$ ($103 \pm 11\%$) for 0.2 mg L^{-1} , $0.507 \pm 0.049 \text{ mg L}^{-1}$ ($105 \pm 8\%$) for 0.5 mg L^{-1} , $1.052 \pm 0.138 \text{ mg L}^{-1}$ ($105 \pm 14\%$) for 1.0 mg L^{-1} , and finally $4.119 \pm 0.404 \text{ mg L}^{-1}$ ($107 \pm 9\%$) for 4.0 mg L^{-1} . Statistical analysis showed no significant differences between these pretreatments. However, the higher recovery suggests a higher and more stable binding affinity of Ba^{2+} to the sulfonic acid groups of the PES membrane as supported by the sequence of retention times by Zenki³⁷ and the above-mentioned affinity series for cations.⁴⁶ Especially for low concentrations of less than 0.2 mg L^{-1} , pretreatment with BaCl_2 could therefore be advantageous for the recovery of copper when using UF for the determination of dissolved Cu.

Due to the toxicity of Ba^{2+} , its consumption should be minimized, and only that quantity of BaCl_2 should be applied that is necessary to occupy all adsorption sites of the PES membrane. For a proof of concept, a method for minimizing BaCl_2 application will be tested.

So far, we tested simple aqueous solutions of Cu^{2+} (as CuSO_4) at neutral pH. For more complex media, influences of the pH, composition of the medium, like ionic strength, complexing components (e.g., EDTA), and content of dissolved organic matter (DOM) are expected. As the main effect for the recovery and the retention of Cu in UF is the Coulomb interaction, we suggest using speciation calculations for the metal cation, focusing on the used test media.

Furthermore, the pH of media, in an ecotoxicological context, is typically in the range of 5–8. In this range, the PES membrane is strongly negatively charged³³ and the speciation of Cu is mainly controlling the interaction with the PES membrane and may be reduced due to the formation of neutral or negatively charged complexes (e.g., increase of the neutral $[\text{Cu}(\text{OH})_2]$ and negatively charged $[\text{Cu}(\text{OH})_3]^-$ species with increasing pH > 8). Additionally, common anions like carbonate CO_3^{2-} or sulfide S^{2-} and the presence of DOM (e.g., humic acid) are known to form complexes and bind strongly to Cu^{2+} .^{17,59} In case the molecular weight of the DOM binding the Cu^{2+} is larger than the MWCO of the membrane, a large part of the Cu^{2+} would be retained by the membrane, like it was shown for, for example, Ag^+ and proteins.¹⁸ The interaction of divalent cations with humic acid and the influence on microfiltration and UF have been discussed in detail.^{60–62}

Also, elevated ionic strength of the medium can increase the retention of especially divalent cations if the pore sizes of the membrane are small and the separation is increasingly controlled by the solution-diffusion mechanism.²⁴ Alternatively, according to the Debye–Hückel theory, elevated ionic strength shields the Coulomb interaction of the cation and the membrane and may reduce the retention by the membrane. Otherwise, UF with a larger pore size and a higher MWCO may lead to higher recovery as reported for the PES membrane with an MWCO of 50 kDa.⁶³

Overall, due to the comparability of the observed retentions in UF experiments with ion exchange experiments, we assume that our approach is well applicable to the speciation of other environmentally relevant metals like zinc, nickel, cobalt, and so forth by UF.

Finally, it is strongly recommended to check the recovery in the concentration range of interest when using separation methods for the determination of the dissolved fraction of metals, especially in the case of UF. Additionally, we strongly suggest performing speciation calculations (e.g., using VMinteq

or PHREEQC)^{64,65} for the metal in focus and the used test media to account for the above discussed influences on the interactions of the membrane and the species of the metal in focus.

CONCLUSIONS

It was shown that the recovery of Cu after UF decreases with decreasing Cu concentration. This is probably due to the Coulomb interaction of the Cu ions with the negatively charged sulfonic acid groups of the PES membrane, similar to retention in small-ion exchange columns.

Assuming that the recovery can be increased by pretreating the membrane to hinder interactions of the PES membrane with the Cu ions, various cations were tested for pretreatment. Ca^{2+} and Ba^{2+} at the concentration 0.1 mol L^{-1} were found to be the most effective divalent cations for pretreatment. This was in line with the affinity series of cations on sulfonated polystyrene cation exchange resins.

For lower Cu concentrations ($\leq 0.2 \text{ mg L}^{-1}$), the pretreatment with 0.1 M CaCl_2 was rather effective concerning the recovery of Cu ($93 \pm 10\%$). However, $0.1 \text{ mol L}^{-1} \text{ BaCl}_2$ could be identified as the most efficient pretreatment ($103 \pm 11\%$). This advantage of Ba ions must be weighed against their high toxicity when used in the laboratory; their use should be avoided whenever possible.

The main purpose of the method establishment was the separation of CuONPs and Cu ions in environmentally relevant Cu concentrations. This was achieved by excellent recovery, especially in the concentration range of 0.2 mg L^{-1} and below. The efficiency for the separation of Cu ions released by CuONPs by pretreatment of the PES membrane and in more complex media is currently being tested.

ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge at <https://pubs.acs.org/doi/10.1021/acsomega.2c07355>.

Details on preliminary experiments, results for additional membrane pretreatments, sample preparation for GF-AAS, method for copper determination by GF-AAS, and data evaluation including error propagation (PDF)

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Notes

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